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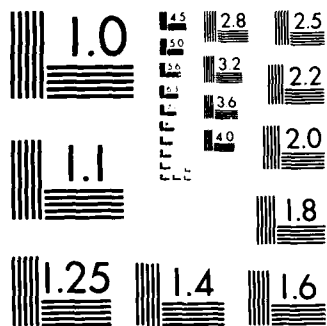
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POLYCYTHEMIA AND HYDRATION STATUS: EFFECTS ON BLOOD VOLUME AND  
THERMOREGULATION DURING EXERCISE-HEAT STRESS

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Michael N. Sawka, Richard R. Gonzales, Andrew J. Young, Stephen R. Muza,  
Kent B. Pandolf, William A. Latzka, Richard C. Dennis and C. Robert Valeri

U.S. Army Research Institute of Environmental Medicine, Natick, MA 01760-  
5007; and Naval Blood Research Laboratory, Boston University School of  
Medicine, Boston, MA 02118

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Abbreviated Title: Polycythemia and Hydration Status

Mailing Address:

Michael N. Sawka, Ph.D.  
U.S. Army Research Institute of  
Environmental Medicine  
Kansas Street  
Natick, MA 01760-5007

DISTRIBUTION STATEMENT A

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SECURITY CLASSIFICATION OF THIS PAGE

A185 946

## REPORT DOCUMENTATION PAGE

Form Approved  
OMB No. 0704-0188  
Exp. Date: Jun 30, 1986

1a. REPORT SECURITY CLASSIFICATION Unclassified			1b. RESTRICTIVE MARKINGS		
2a. SECURITY CLASSIFICATION AUTHORITY			3. DISTRIBUTION / AVAILABILITY OF REPORT Approved for public release; distribution is unlimited.		
2b. DECLASSIFICATION / DOWNGRADING SCHEDULE			5. MONITORING ORGANIZATION REPORT NUMBER(S)		
4. PERFORMING ORGANIZATION REPORT NUMBER(S)			7a. NAME OF MONITORING ORGANIZATION U.S. Army Research Institute of Environmental Medicine		
6a. NAME OF PERFORMING ORGANIZATION U.S. Army Res Inst of Env Med		6b. OFFICE SYMBOL (if applicable) SGRD-UE-MEP	7b. ADDRESS (City, State, and ZIP Code) Kansas Street Natick, MA 01760-5007		
6c. ADDRESS (City, State, and ZIP Code) Kansas Street Natick, MA 01760-5007			7c. ADDRESS (City, State, and ZIP Code) Kansas Street Natick, MA 01760-5007		
8a. NAME OF FUNDING / SPONSORING ORGANIZATION Same as 6a.		8b. OFFICE SYMBOL (if applicable)	9. PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER		
8c. ADDRESS (City, State, and ZIP Code) Same as 6c.			10. SOURCE OF FUNDING NUMBERS		
			PROGRAM ELEMENT NO. A879	PROJECT NO. 3E162777 A879	TASK NO. 879/BD
			WORK UNIT ACCESSION NO. 127		
11. TITLE (Include Security Classification) (U) Polycythemia and Hydration Status: Effects on Blood Volume and Thermoregulation During Exercise-Heat Stress					
12. PERSONAL AUTHOR(S) Michael N. Sawka, Richard R. Gonzalez, Andrew J. Young, Stephen R. Muza, Kent B. Pandolf, William A. Latzka, Richard C. Dennis and C. Robert Valeri					
13a. TYPE OF REPORT Manuscript		13b. TIME COVERED FROM _____ TO _____		14. DATE OF REPORT (Year, Month, Day) July 1987	
				15. PAGE COUNT 19	
16. SUPPLEMENTARY NOTATION					
17. COSATI CODES			18. SUBJECT TERMS (Continue on reverse if necessary and identify by block number)		
FIELD	GROUP	SUB-GROUP	blood "doping", blood reinfusion, dehydration, erythrocythemia, euhydration, heat acclimation, hypohydration, plasma volume, red blood cells, temperature regulation, transfusion		
19. ABSTRACT (Continue on reverse if necessary and identify by block number) We studied the effects of autologous erythrocyte reinfusion on blood volume and thermoregulation during exercise in the heat. Five heat-acclimated males attempted four Heat Stress Tests (HSTs): two pre- and two post-reinfusion. Autologous erythrocyte reinfusion was accomplished with 500 ml of a NaCl-glucose-phosphate solution containing 60% hematocrit. Both pre- and post-reinfusion one HST was done while euhydrated, and one HST was done while hypohydrated (~5% of body weight). After 30 min of rest in a 20°C antechamber, the HST consisted of a 120-min exposure (2 repeats of 25 min rest and 45 min walking) in a hot (35°C, 45% rh) environment. The following new findings were made concerning acute polycythemia in heat-acclimated subjects: 1) the increased erythrocyte volume was associated with a small plasma volume expansion; 2) the plasma volume expansion was associated with an increased total circulating protein mass; 3) the increased total circulating protein mass defends plasma volume when hypohydrated; 4) polycythemia increased sweating rate and reduced core temperature during exercise-heat stress; and 5) this thermoregulatory					
20. DISTRIBUTION / AVAILABILITY OF ABSTRACT <input checked="" type="checkbox"/> UNCLASSIFIED/UNLIMITED <input type="checkbox"/> SAME AS RPT. <input type="checkbox"/> DTIC USERS			21. ABSTRACT SECURITY CLASSIFICATION		
22a. NAME OF RESPONSIBLE INDIVIDUAL Dr. Michael N. Sawka			22b. TELEPHONE (Include Area Code) 617-651-5141		22c. OFFICE SYMBOL SGRD-UE-MEP

DD FORM 1473, 84 MAR

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All other editions are obsolete.SECURITY CLASSIFICATION OF THIS PAGE  
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# ABSTRACT

We studied the effects of autologous erythrocyte reinfusion on blood volume and thermoregulation during exercise in the heat. Five heat-acclimated males attempted four Heat Stress Tests (HSTs): two pre- and two post-reinfusion. Autologous erythrocyte reinfusion was accomplished with 500 ml of a NaCl-glucose-phosphate solution containing ~80% hematocrit. Both pre- and post-reinfusion one HST was done while euhydrated and one HST was done while hypohydrated (-5% of body weight). After 30 min of rest in a 20°C antechamber, the HST consisted of a 120-min exposure (2 repeats of 15 min rest and 45 min walking) in a hot (35°C, 45% rh) environment. The following new findings were made concerning acute polycythemia in heat-acclimated subjects: (1) the increased erythrocyte volume was associated with a small plasma volume expansion; (2) the plasma volume expansion was associated with an increased total circulating protein mass; (3) the increased total circulating protein mass defends plasma volume when hypohydrated; (4) polycythemia increased sweating rate and reduced core temperature during exercise-heat stress; and (5) this thermoregulatory advantage conferred by acute polycythemia was effective even during hypohydration. Additionally, observations were made providing evidence that heat acclimation may increase extravascular protein mass.

Index Terms: blood "doping", blood reinfusion, dehydration, erythrocythemia, euhydration, heat acclimation, hypohydration, plasma volume, red blood cells, temperature regulation, transfusion



Classification	
Distribution/	
Availability Codes	
Avail and/or	
Special	
A-1	

## INTRODUCTION

Recently, we found that acute polycythemia reduced heat storage during exercise in a hot environment (14). That investigation employed a group of euhydrated but non-heat acclimated subjects. When infused with an autologous erythrocyte-saline solution, those subjects had a reduced plasma volume so that they maintained the same blood volume as during the pre-reinfusion measurements. The reduced plasma volume at rest and during exercise was associated with a reduced total circulating protein mass. Again, despite the reduced plasma volume, but same blood volume, those subjects still possessed a small thermoregulatory advantage during exercise in the heat.

Our previous study raised several questions concerning the use of acute polycythemia as an ergogenic aid during exercise in the heat. First, will the small thermoregulatory advantage conferred by acute polycythemia still be present in heat acclimated subjects? Heat acclimation enables an individual to perform exercise in the heat with reduced heat storage, and may elicit optimal thermoregulatory benefits that acute polycythemia cannot improve upon. Second, if heat acclimated subjects receive an autologous erythrocyte reinfusion, will they also demonstrate a compensatory plasma volume reduction to maintain the same blood volume? Heat acclimated subjects possibly have a greater availability of extravascular protein which might be "washed" into the intravascular space by the reinfusion and allow a plasma volume defense or expansion with acute polycythemia. Third, will acute polycythemia provide a thermoregulatory advantage or disadvantage in hypohydrated subjects during exercise in the heat? Hypohydration will reduce plasma volume, and this reduction may be accentuated by the acute polycythemia and could provide a thermoregulatory disadvantage during subsequent exercise in the heat.

In the present study, we examined the effects of autologous erythrocyte reinfusion on blood volume and thermoregulation during exercise in the heat. These experiments were performed on heat acclimated subjects when both euhydrated and hypohydrated.

## METHODS

Subjects. Five fit male volunteers participated in this investigation. Four additional subjects volunteered and had phlebotomies, but were unavailable for testing. The subjects ( $n=5$ ) had a mean ( $\pm$ SD) age of  $33\pm 2$  yr, weight of  $86\pm 13$  kg, surface area-to-mass ratio of  $234\pm 14$   $\text{cm}^2\cdot\text{kg}^{-1}$ , and percent body fat of  $20\pm 5$ .

Protocol. During the spring-summer months, two units of blood were removed by phlebotomy from each subject. A minimum of six weeks separated the removal of each unit of blood. During the subsequent fall-early winter months, the experimental portion of the study was completed. Initially, the subjects were familiarized with the test procedures, had their percent body fat determined by hydrostatic weighing, and completed a maximal aerobic power test. In addition, on fifteen days during the three weeks prior to experimental testing and throughout the study, nude body weights were measured in the morning after voiding and before breakfast. These body weights were used to establish base line body weights that represented euhydration for each subject.

Subjects were heat acclimated by walking on a treadmill (0-6% grade) at  $1.34 \text{ m}\cdot\text{s}^{-1}$  for two 50-minute exercise bouts separated by a 10-minute rest period for nine days in a hot-dry ( $45^\circ\text{C}$  ambient temperature, 20% relative humidity) environment. In addition, heat acclimation sessions were completed on the days between the experimental Heat Stress Tests (ESTs). During all



tests, subjects wore gym shorts, and tennis shoes; the subjects drank water ad libitum during the acclimation sessions. Following the nine-day heat acclimation program, the subjects had their resting plasma volume measured on one day and then on other days completed two HSTs before erythrocyte reinfusion. One HST test was done while euhydrated and the other HST test was done while hypohydrated by 5% of body weight. Approximately two days after completing the second pre-reinfusion HST, each subject received an autologous erythrocyte reinfusion. They received ~500 ml of a sodium-chloride-glucose-phosphate solution containing ~60% hematocrit (autologous erythrocytes). Plasma volume was measured 24 h post-reinfusion, and two HST's (euhydrated and hypohydrated) were done 48 h and 96 h post-reinfusion. A test of maximal aerobic power was completed 72 h post-reinfusion. The order of the pre-infusion tests were counterbalanced; however, post-reinfusion logistics required each subject to first do the euhydration HST and then do the hypohydration HST.

Phlebotomy, blood storage, reinfusion and plasma volume measurements were conducted at the Naval Blood Research Laboratory. After each phlebotomy, the blood was separated into its erythrocyte and plasma components, and the erythrocytes were frozen with 40% (wt/vol) glycerol and stored at -80°C (22,23). For reinfusion ~500 ml of autologous erythrocytes in a sodium chloride-glucose-phosphate solution were administered over a 1-h period. The frozen cell component was thawed and washed to reduce the glycerol concentration to <1%. The erythrocyte O<sub>2</sub> transport function was determined from the in vitro state reported as the P<sub>50</sub> value, which is the partial pressure at which the hemoglobin is 50% saturated (3).

The maximal aerobic power and HSTs were conducted at the U.S. Army Research Institute of Environmental Medicine. Each subject's maximal aerobic

power was determined by a progressive-intensity continuous-effort treadmill test. The warm-up bout consisted of 10 min of walking ( $1.56 \text{ m}\cdot\text{s}^{-1}$ ) at a 10% treadmill grade. The subjects then ran ( $2.68$  or  $3.13 \text{ m}\cdot\text{s}^{-1}$ ) continuously at an initial grade of 5% with 2.5% increments every 1.5 minutes. Established criteria were employed for determination of maximal  $\text{O}_2$  uptake (18,21). These tests were conducted in a comfortable ( $20^\circ\text{C}$  ambient temperature, 40% relative humidity) environment.

The HSTs were conducted in a hot ( $35^\circ\text{C}$  ambient temperature, 45% relative humidity) environment. This environment was selected to enable limited sensible and potentiate insensible heat exchange. Each HST was 120 min (2 repeats of 15 min rest and 45 min exercise) in duration. During exercise, subjects walked on an inclined treadmill, and during the rest periods they were weighed and rehydrated with spring water to maintain their desired body weight (i.e., euhydration or hypohydration level).

Approximately 24-48 h prior to each hypohydration HST, subjects initiated a program of voluntary food and fluid restriction. Also, on the afternoon prior to each HST, subjects performed light-intensity exercise in a hot ( $T_a = 45^\circ\text{C}$ ) environment to dehydrate to their target body weight (5% below baseline). After achieving the target body weight, subjects were removed to a comfortable environment to spend the night. During the night, subjects were allowed fresh fruit and juice but only in amounts that their weight allowed. In addition, as a control the subjects performed exercise in this hot environment with adequate rehydration prior to the euhydration HSTs. All subjects completed the dehydration 15-18 h prior to the HSTs. The following morning the subjects were weighed (0700 h), provided fresh fruit and juice (as weight allowed), instrumented and tested (0930 h).

Measurements. Electrocardiogram was obtained with chest electrodes (CM5 placement) and radiotelemetered to an oscilloscope-cardiotachometer unit (Hewlett-Packard). During the maximal aerobic power tests, an automated system (Sensormedics Horizon MMC) was used to measure  $O_2$  uptake. During the HSTs, the respiratory gases were collected in 150-liter Douglas bags. The volume of expired gases was measured with a Tissot gasometer, and the  $O_2$  and  $CO_2$  concentrations were measured with an electrochemical  $O_2$  analyzer (Applied Electrochemistry S-3A) and an infrared  $CO_2$  analyzer (Beckman LB-2), respectively.

During the HSTs, rectal temperature was measured from a thermistor inserted ~10 cm beyond the anal sphincter. Skin temperatures were obtained with a three-point thermocouple skin harness (chest, calf, and forearm), and mean weighted skin temperature ( $T_{sk}$ ) was calculated (1). The dew point temperature of the upper arm was continuously measured by a ventilated sensor and local sweating rate was calculated (8). Total body sweating rates were calculated from nude body weight loss adjusted for water intake and urine output.

Venous blood samples were collected from an indwelling Teflon catheter placed within a superficial forearm vein. Patency was maintained with heparinized saline; the catheter (2 ml of dead space) was flushed with 4 ml of blood before each 8 ml sample was obtained. Blood samples taken at rest were obtained while all the subjects stood (for 20 min prior to sampling) in the antechamber (20°C ambient temperature, 40% relative humidity), and exercise blood samples were obtained 15 min and 35 min into each exercise bout while the subjects continued to walk. Triplicate measurements were made for all blood variables. Automated systems were used to measure hemoglobin (Hemoglobinometer, Coulter Electronics) and plasma lactate (Model 23 lactate

analyser, Yellow Springs Instrument). Plasma osmolality was measured by a vapor pressure osmometer (Model 5500, Wescor) and plasma protein content was quantified with a refractometer (American Optical). Plasma volume at rest (euhydrated) was measured by the iodine-labeled ( $^{125}\text{I}$ ) albumin method and erythrocyte volume was calculated from the measured plasma volume and total body hematocrit (24). The percent change in plasma volume was calculated from the appropriate hemoglobin and hematocrit values (4). The plasma volumes during exercise were calculated by adjusting the measured plasma volume at rest by the appropriate percent change in plasma volume. Blood volume was calculated as the sum of plasma volume and erythrocyte volume. Total circulating protein mass was calculated as the product of plasma volume and plasma protein content.

Statistical Analysis. Mean  $\pm$  SD, simple regression, and repeated measures analyses of variance followed by Tukey's post hoc procedures were used.

## RESULTS

Acclimation. After a general decrease from the values on the initial day, final exercise rectal temperature ( $T_{re}$ ) and heart rate responses did not change during the final five heat acclimation days, indicating complete heat acclimation. Also, rectal temperature and heart rate responses were nearly identical during each additional heat acclimation day conducted between HSTs.

Infusion. For each subject, resting erythrocyte volume, hemoglobin concentration and venous hematocrit increased after erythrocyte reinfusion. For the group, erythrocyte volume went from  $1.97 \pm 0.26$  to  $2.16 \pm 0.23$  liters ( $p > 0.05$ ), hemoglobin increased ( $p < 0.10$ ) from  $14.9 \pm 0.7$  to  $15.8 \pm 1.0 \text{ g}\cdot\text{dl}^{-1}$  and venous hematocrit ( $p < 0.05$ ) increased from  $44.6 \pm 1.9$  to  $47.4 \pm 2.2$  divisions. The erythrocyte  $P_{50}$  value was  $27 \pm 1$  and  $28 \pm 1$  Torr pre- and post-reinfusion, respectively.

Maximal Exercise. For each subject, maximal aerobic power increased after erythrocyte reinfusion. One subject discontinued the post-reinfusion maximal exercise test prematurely, without achieving the criterion for maximal aerobic power, due to aggravation of an old back injury. For the group, maximal aerobic power values were  $4.26 \pm 0.23$  and  $4.48 \pm 0.30 \text{ l} \cdot \text{min}^{-1}$  ( $p > 0.05$ ) pre- and post-reinfusion, respectively.

Hypohydration. During the hypohydration experiments, the subjects had a mean body weight of 81.7 kg, which corresponded to  $5.0 \pm 0.0\%$  below their baseline body weights. For resting subjects, hypohydration increased ( $p < 0.01$ ) plasma osmolality (Table 1). The percent change in resting plasma volume from euhydration to hypohydration levels was  $-13 \pm 6$  (range -5 to -22%) for the pre-reinfusion and  $-4 \pm 9$  (range +5 to -16%) for the post-reinfusion hypohydration experiments. Figure 1A provides the individual data showing a tendency for a smaller ( $p < 0.10$ ) reduction in plasma volume when hypohydrated (compared to when euhydrated) after erythrocyte reinfusion. Figure 1B indicates that the one subject (#5) who did not defend his plasma volume also failed to increase his resting total circulating protein mass during the post-reinfusion measurements. During the HSTs, hypohydration elevated plasma osmolality ( $p < 0.01$ ), but did not alter plasma volume or plasma lactate from euhydration levels (Table 1). In addition, hypohydration increased rectal temperature ( $p < 0.05$ ), heart rate ( $p < 0.05$ ) and decreased total body sweating rate ( $p < 0.05$ ) compared to the euhydration HST levels (Table 2).

Heat Stress Tests, Reinfusion. All five subjects completed (120 min) both euhydration HSTs, however, during the hypohydration HSTs only three subjects completed both experiments. The other two subjects demonstrated an increased endurance when hypohydrated during the post- compared to pre-reinfusion HSTs. One subject increased his endurance time from 40 to 87

minutes and the other subject increased his endurance time from 60 to 79 minutes from the pre- to post-reinfusion hypohydration HSTs.

Table 1 provides the subjects' hematological measurements during the four HSTs. Erythrocyte reinfusion resulted in reduced ( $p < 0.05$ ) plasma osmolality, due to only the hypohydration experiments. Plasma volume ( $p < 0.05$ ) and total circulating protein mass ( $p < 0.05$ ) were increased by erythrocyte reinfusion, but plasma lactate was not altered. Also, the plasma protein content ( $\text{g} \cdot \text{dl}^{-1}$ ) was not altered by erythrocyte reinfusion. Figure 3 depicts the subjects' blood volumes during the four HSTs. Note that erythrocyte reinfusion increased ( $p < 0.01$ ) blood volume, and that the pre-reinfusion euhydration volumes and post-reinfusion hypohydration volumes were nearly identical.

Table 2 provides the subjects' metabolic rate and thermoregulatory measurements during the four HSTs. Erythrocyte reinfusion did not alter metabolic rate during exercise which approximated  $45 \pm 6\%$  of their initial maximal aerobic power. During exercise the rectal temperature values were reduced ( $p < 0.01$ ) post-reinfusion, and the individual final exercise values are presented in Figure 2A. The sweating rate data presented in Table 2 were only for the first exercise bout, as subject attrition and measurement problems resulted in too many missing data points for analyses of the second exercise bout. The onset time for the initiation of thermoregulatory sweating tended ( $p = 0.06$ ) to be lower post-reinfusion, and the individual data are presented in Figure 2B. Both steady-state local sweating rate ( $p < 0.01$ , Figure 2C) and total body sweating rate ( $p < 0.05$ ) were greater post-reinfusion. The mean skin temperatures averaged  $34.0 \pm 0.6$  and  $34.0 \pm 0.7^\circ\text{C}$  during exercise for the pre- and post reinfusion HSTs, respectively.

Figure 4 presents the individual data for the subjects' final rectal temperature difference (top) and (bottom) steady-state local sweating rate

difference (post minus pre-reinfusion) plotted against their blood volume difference and plasma osmolality difference (post- minus pre-reinfusion) during exercise. Significant relationships were not found between any of these variables.

Table 3 presents the subjects' heart rate and perceptual measurements during rest and exercise in the heat. Erythrocyte reinfusion resulted in reduced ( $p < 0.01$ ) heart rate responses. Erythrocyte reinfusion reduced thermal sensation ( $p < 0.01$ ), and tended to lower central ( $p = 0.06$ ) and overall ( $p < 0.10$ ) rated perceived exertion.

## DISCUSSION

We do not advocate the use of blood reinfusions or "blood doping" as an ergogenic aid for athletic competition, as its use for this purpose is unsanctioned and unethical. Our interest in the potential use of erythrocyte reinfusion is for special military situations where it might improve the probability of successfully completing a mission and reducing the number of heat casualties. Finally, erythrocyte reinfusion provides a powerful tool to further our understanding of physiological control mechanisms in response to exercise-heat stress.

The experimental design dictated that each subject complete all four HSTs and serve as his own control. These experiments were conducted after completion of a heat acclimation program so that experimental order (reinfusion following the control HSTs) should not have biased the results. The exercise intensity (40-50%  $\dot{V}O_2$  max) was selected to represent that frequently associated with sustained self-paced work. Hypohydration was achieved with voluntary food and fluid denial combined with physical exercise in a hot environment. A period of 15-18 h was spent resting in a comfortable

environment (while hypohydrated) to provide time for fluid compartments to equilibrate at the achieved hydration level. These dehydration procedures are consistent with those of previous investigations (2,9,16,18).

Blood volume was substantially increased during both rest and exercise after erythrocyte reinfusion. The increase was the sum of the small plasma volume expansion and the additional erythrocytes. The small plasma volume expansion was oncotically mediated as it was associated with a substantial (~20 gram) increase of total circulating protein mass. This amount of protein will exert sufficient oncotic pressure to retain the additional fluid within the intravascular space (19). Erythrocytes do not exert an in vitro oncotic pressure, but may exert an in vivo oncotic pressure (25). The observation of plasma volume defense or slight expansion after erythrocyte reinfusion in heat-acclimated subjects is striking because previously we reported (14) that non-heat acclimated subjects had a decrease in total circulating protein mass and plasma volume after erythrocyte reinfusion. In addition, it is generally accepted that the infusion of extra erythrocytes will result in a compensatory reduction of plasma volume to maintain the pre-infusion blood volume (10). Therefore, mechanisms inherent in the heat acclimation process may modify a subject's total circulating protein mass and plasma volume response to erythrocyte reinfusion.

Heat acclimation is associated with an increased amount of intravascular protein (19); perhaps extravascular protein is also increased and the infusion "washed" these additional extravascular proteins into the intravascular space where they were retained. Consistent with our findings, Valeri and Altschule (24) reported that erythrocyte transfusion increased plasma volume and intravascular albumin in trauma patients who had been transported from Southeast Asia during the preceding two weeks. These individuals were



probably heat acclimated from living in a warm environment. These investigators have found similar results in an animal study that was primarily conducted in the summer months in a warm-humid vivarium (25). Possibly then, heat acclimation alters protein and plasma volume responses to erythrocyte reinfusion.

Erythrocyte reinfusion tended to result in a better defense of resting plasma volume when the subjects were hypohydrated. The smaller plasma volume reduction when hypohydrated was mediated by the increased total circulating protein mass. The four subjects who defended their resting plasma volume when hypohydrated also increased their total circulating protein mass post-reinfusion, whereas subject #5 who did not defend his plasma volume also did not increase his total circulating protein mass (Figure 1B). The question arises as to why erythrocyte reinfusion did not increase that subject's total circulating protein mass. We believe that the reason resides in his large body mass and plasma volume, as he was by far our largest subject weighing 108 kg and having a resting plasma volume of ~3.7L. All subjects were reinfused with the same volume of erythrocyte in saline solution. For this large subject, the reinfusion presented the smallest volume overload to the circulatory system, so that less infused fluid was filtered from the intravascular space through a given interstitial space (to "washout" protein) and translocate protein via lymph to the intravascular spaces. This suggests that the volume of saline solution reinfused with the erythrocytes may influence the elevation of total circulating protein mass in heat-acclimated subjects.

Erythrocyte reinfusion provides a thermoregulatory advantage to subjects exercising in the heat regardless of their hydration status. This thermoregulatory advantage was demonstrated by both a reduced core temperature

and improved sweating rate during exercise-heat stress. Previously, we found that acute polycythemia provided a small thermoregulatory advantage for euhydrated but non-heat acclimated subjects (14). The present study has both confirmed and expanded upon those findings. If anything, acute polycythemia provided a more pronounced thermoregulatory advantage in the heat acclimated subjects. It is known that heat acclimation improves both insensible and sensible heat loss to reduce core temperature during exercise-heat stress (2,16). Perhaps, acute polycythemia is able to take better advantage of the improved thermoregulatory effector responses of heat acclimated individuals.

The physiological mechanisms responsible for the thermoregulatory advantage provided by acute polycythemia are probably different for the euhydration and hypohydration experiments. These mechanisms might include: (a) an increased blood volume; (b) a reduced plasma osmolality; and (c) an increased total body water. A defense of blood volume would clearly mediate a thermoregulatory advantage in the hypohydration experiments but perhaps not in the euhydration experiments. Hypohydration decreases an individual's blood volume, resulting in impaired insensible and sensible heat loss, thus elevating core temperature in comparison to euhydration (7,13,16,17). This thermoregulatory disadvantage from dehydration is somewhat related to the magnitude of hypovolemia (5,6,13,17) and can be reversed by the re-establishment of the normal blood volume (20) during exercise in the heat. Therefore, during the post-reinfusion hypohydration experiments, the maintenance of blood volume at essentially normovolemic levels would mediate a thermoregulatory advantage compared to the pre-reinfusion hypohydration HST where hypovolemia occurred.

The thermoregulatory advantages mediated by an increased blood volume are less clear-cut for the euhydration experiments. Several investigators report

that hyperhydration and/or hypervolemia do not provide a thermoregulatory advantage over euhydration and/or normovolemia (5,9,13,15). Moroff and Bass (12) examined the influence of excessive fluid ingestion on thermoregulatory responses to exercise in the heat. They reported that hyperhydration reduced core temperature while elevating sweating rate above euhydration level. During their euhydration experiments, however, their subjects experienced a slight progressive dehydration. Therefore, those results may demonstrate the effects of hypohydration rather than hyperhydration. Three studies have artificially expanded blood volume and found that acute hypervolemia did not provide a thermoregulatory advantage, compared to normovolemia, during exercise in the heat (5,13,15). In contrast, Fortney et al. (6) found that acute hypervolemia lowered core temperature below normovolemia levels during exercise in a hot environment. This was observed despite no difference in the sweating rate in a 30°C environment. Therefore, it is possible that hypervolemia may have contributed to the thermoregulatory advantage in the post-reinfusion euhydration experiments.

Plasma hyperosmolality has been shown to reduce thermoregulatory effector responses and thus elevate core temperature during exercise in the heat (7,17). During the euhydration experiments, plasma osmolality values were not different between the pre- and post-reinfusion HSTs. During the hypohydration experiments, however, plasma osmolality values were significantly lower post-reinfusion. These lower osmolality values might be explained by a larger plasma volume or total body water. The subjects dehydrated by losing the same amount of body weight via sweat secretion both pre- and post-reinfusion. Eccrine sweat is hypotonic relative to plasma, therefore, plasma will become hypertonic when hypohydration is induced by sweat output (16,17). If an equal number of solutes were retained in the plasma when hypohydrated both pre- and

post-reinfusion, a lower plasma osmolality would indicate a larger volume of distribution. This larger volume of distribution could be an expanded plasma volume or total body water. Regardless, maintenance of a lower plasma osmolality following reinfusion when hypohydrated provides a mechanism to mediate improved thermoregulatory effector responses.

In this study we did not perform the measurements to quantitate sensible heat loss as the selected environment kept radiative and convective heat exchange to a minimum. As stated above, a smaller blood volume reduction and reduced hyperosmolality can both mediate improved thermoregulatory responses for insensible as well as sensible heat exchange (7,17). We can hypothesize an additional physiological mechanism for improved sensible heat exchange after erythrocyte reinfusion: a reduced skeletal muscle blood flow may allow increased cutaneous blood flow at a given cardiac output during submaximal exercise. Several human and animal studies report that hyperoxia will reduce skeletal muscle blood flow during submaximal exercise (26,27,28).

Our data indicate several new findings concerning acute polycythemia in heat-acclimated subjects: 1) the increased erythrocyte volume was associated with a small plasma volume expansion; 2) the plasma volume expansion was associated with an increased total circulating protein mass; 3) the increased total circulating protein mass will defend plasma volume when hypohydrated; 4) polycythemia will increase sweating rate and reduce core temperature during exercise-heat stress; and 5) the thermoregulatory advantage conferred by acute polycythemia was effective even during hypohydration. Additionally, observations were made providing evidence that heat acclimation may increase extravascular protein mass.

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## ACKNOWLEDGEMENTS

Recognition is due to Ron Carciere, George Cassidy, Holly Feingold, Alan Gray, Linda Pivacek and Gina Ragno for the collection and processing of the blood for storage. The authors gratefully acknowledge James Bogart, Bruce Cadarette, Leslie Levine, Darrell Neuffer and Karen Speckman for technical assistance, Patricia DeMuis in preparing the manuscript and Robert Oster for statistical analysis.

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Human subjects participated in these studies after giving their free and informed voluntary consent. Investigators adhered to AR 70-25 and USAMRDC Regulation 70-25 on Use of Volunteers in Research.

This study was supported in part by the Office of Navy (ONR Contract N00014-79-0168) with the funds provided by the Naval Medical Research and Development Command.



Table 1. Influence of erythrocyte reinfusion on hematological measurements during rest and exercise in the heat.

n=5		Osmolality (mosmol $\cdot$ kg $^{-1}$ )		Plasma Volume (L)		Total Circulating Protein (g)		Lactate (mmol $\cdot$ L $^{-1}$ )	
		Rest	Ex-1	Ex-2	Rest	Ex-1	Ex-2	Rest	Ex-2
<b>Euhydrated</b>									
<b>Pre-Reinfusion</b>									
$\bar{X}$		283	287	286	3.38	3.54	3.52	278	281
SD		3	2	4	0.35	0.43	0.37	40	38
<b>Post-Reinfusion</b>									
$\bar{X}$		283	284	284	3.46	3.66	3.74	297	296
SD		1	2	2	0.48	0.47	0.48	37	38
<b>Hypohydrated</b>									
<b>Pre-Reinfusion</b>									
$\bar{X}$		297	300	301	3.00	3.31	3.14	282	264
SD		5	4	3	0.36	0.18	0.10	45	6
<b>Post-Reinfusion</b>									
$\bar{X}$		293	293	298	3.35	3.44	3.31	300	280
SD		6	5	3	0.29	0.2	0.15	37	15
								2.0	1.1
								0.7	0.3

Ex-1 and Ex-2 are the first and second exercise bouts, respectively. For the hypohydration experiments, the Ex-2 values represent only three subjects.

Table 2. Influence of erythrocyte reinfusion on thermoregulatory measurements during exercise-heat stress.

n=5	Metabolic Rate <sub>2</sub> (W·m <sup>-2</sup> )	Final Rectal Temperature (°C)	Local Sweating Onset Time (min)	Steady-State		Total Body Sweating Rate (g·m <sup>-2</sup> ·h <sup>-1</sup> )
				Local Sweating Rate <sub>2</sub> (mg·cm <sup>-2</sup> ·min <sup>-1</sup> )	Rate <sub>2</sub> (mg·cm <sup>-2</sup> ·min <sup>-1</sup> )	
Euhydrated						
Pre-Reinfusion						
X	322	38.0	5.7	0.56		454
SD	22	0.4	2.6	0.24		62
Post-Reinfusion						
X	325	37.8	3.3	0.85		507
SD	20	0.3	1.9	0.21		33
Hypohydrated						
Pre-Reinfusion						
X	325	38.9	5.4	0.62		411
SD	20	0.4	2.6	0.13		61
Post-Reinfusion						
X	319	38.5	1.9	0.87		445
SD	18	0.5	0.4	0.23		48

For the two subjects who did not complete the entire hypohydration HSTs, the hypohydration values are standardized for the value at the time when they discontinued the pre-reinfusion hypohydration test.

Table 3. Influence of erythrocyte reinfusion on heart rate and perceptual measurements during rest and exercise in the heat

	Heart Rate (bpm)			Ratings of Perceived Exertion						Thermal Sensation			
	Rest	Ex-1	Ex-2	L	C		O	L	C	O	Ex-1	Ex-2	
					Ex-1	Ex-2							
n = 5													
<hr/>													
Euhydrated													
Pre-Reinfusion			138	12	11	11	13	12	12	12	5	5	5
X	76	129	22	1	1	1	2	1	1	1	1	1	1
SD	8	14											
Post-Reinfusion			130	9	10	10	12	11	11	11	4	4	4
X	61	125	18	5	2	1	1	1	1	1	1	1	1
SD	5	20											
<hr/>													
Hypohydrated													
Pre-Reinfusion			160	14	13	13	15	13	14	14	6	6	6
X	97	151	18	3	3	3	3	2	2	2	1	1	1
SD	14	11											
Post-Reinfusion			150	14	12	13	13	12	12	12	5	5	5
X	83	150	17	2	2	2	1	1	1	1	1	1	1
SD	6	19											

Ex-1 and Ex-2 are the first and second exercise bouts, respectively. For the hypohydration experiments, the Ex-2 values represent only three subjects. L, C and O represent local, central and overall perceived exertion, respectively.

## FIGURE LEGENDS

Figure 1. Individual data for the percent change in resting plasma volume from euhydration levels to hypohydration levels when the subject has a five percent reduction in body weight (1A). In the figure 1B, the individual relationships between the plasma volume defense and increased total circulating protein mass are plotted. The dotted line represents the theoretical relationship between these variables if only oncotic forces maintain the plasma volume defense (18).

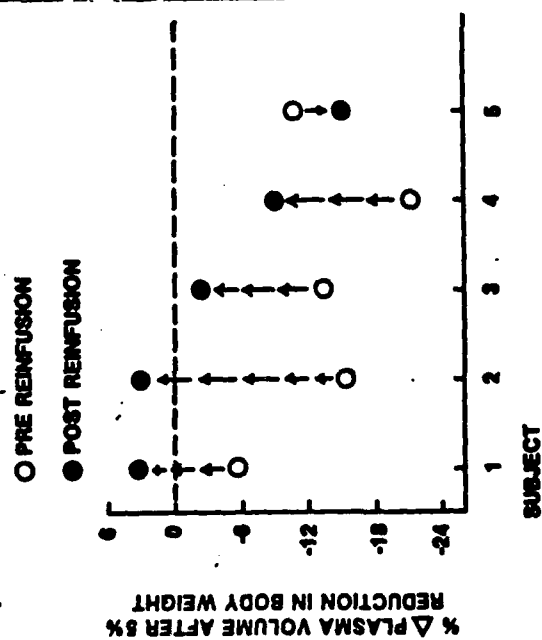
Figure 2. Individual data for the (A) final exercise rectal temperature, (B) local sweating onset time, and (C) steady-state local sweating responses to the pre- and post-reinfusion HSTs.

Figure 3. Blood volume responses at rest and exercise during the four experimental conditions.

Figure 4. The individual data for the subjects' final rectal temperature differences (post- minus pre-reinfusion) plotted against their blood volume ( $r = -0.29$ ) and plasma osmolality ( $r = 0.52$ ) difference (top). Also, individual data for the steady-state local sweating difference plotted against their blood volume ( $r = -0.47$ ) and plasma osmolality ( $r = 0.04$ ) difference (bottom).

Fig. 1

A



B

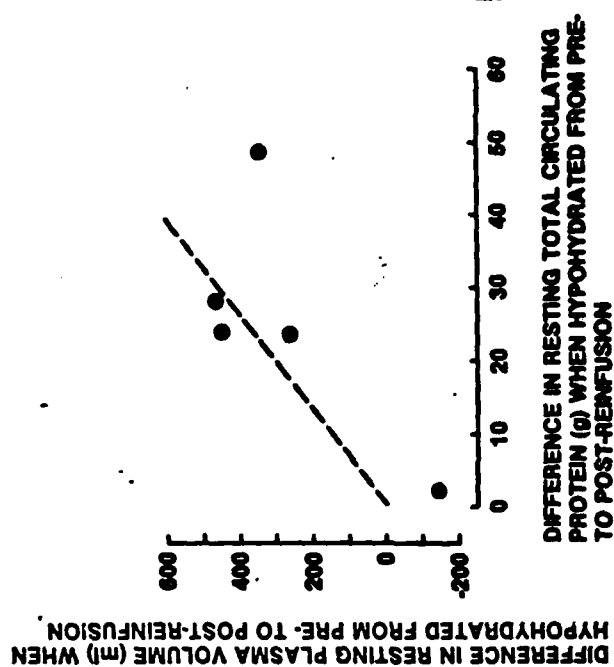
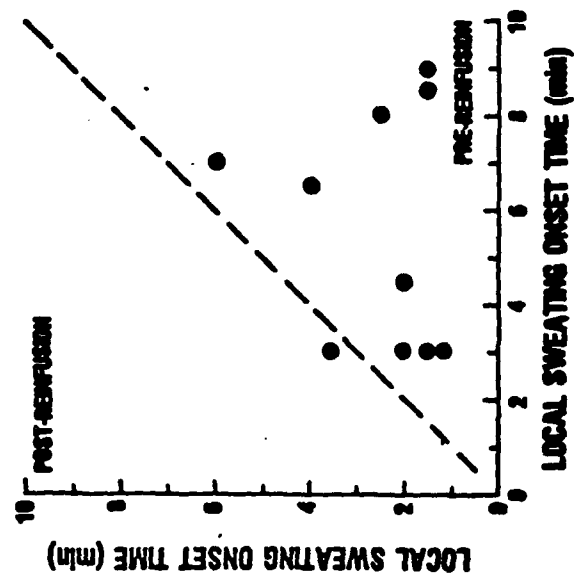
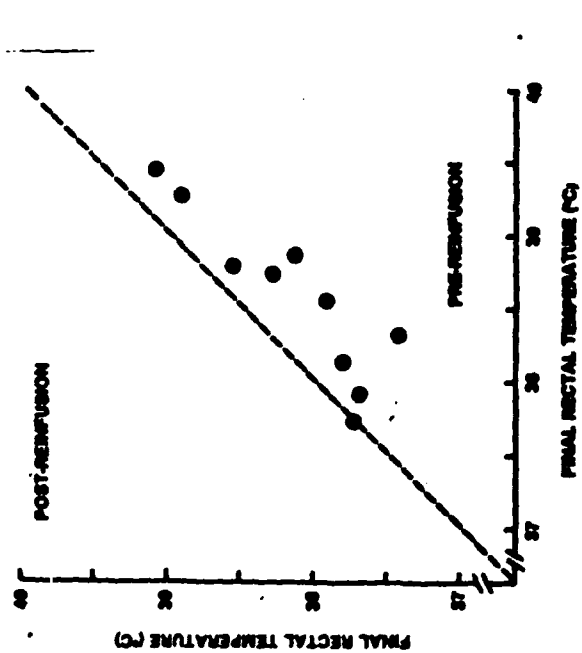


Fig. 2

A



C

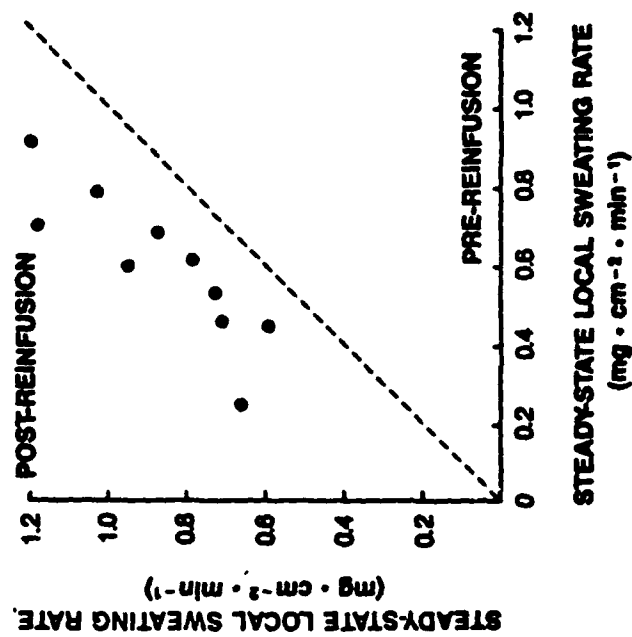


Fig. 3

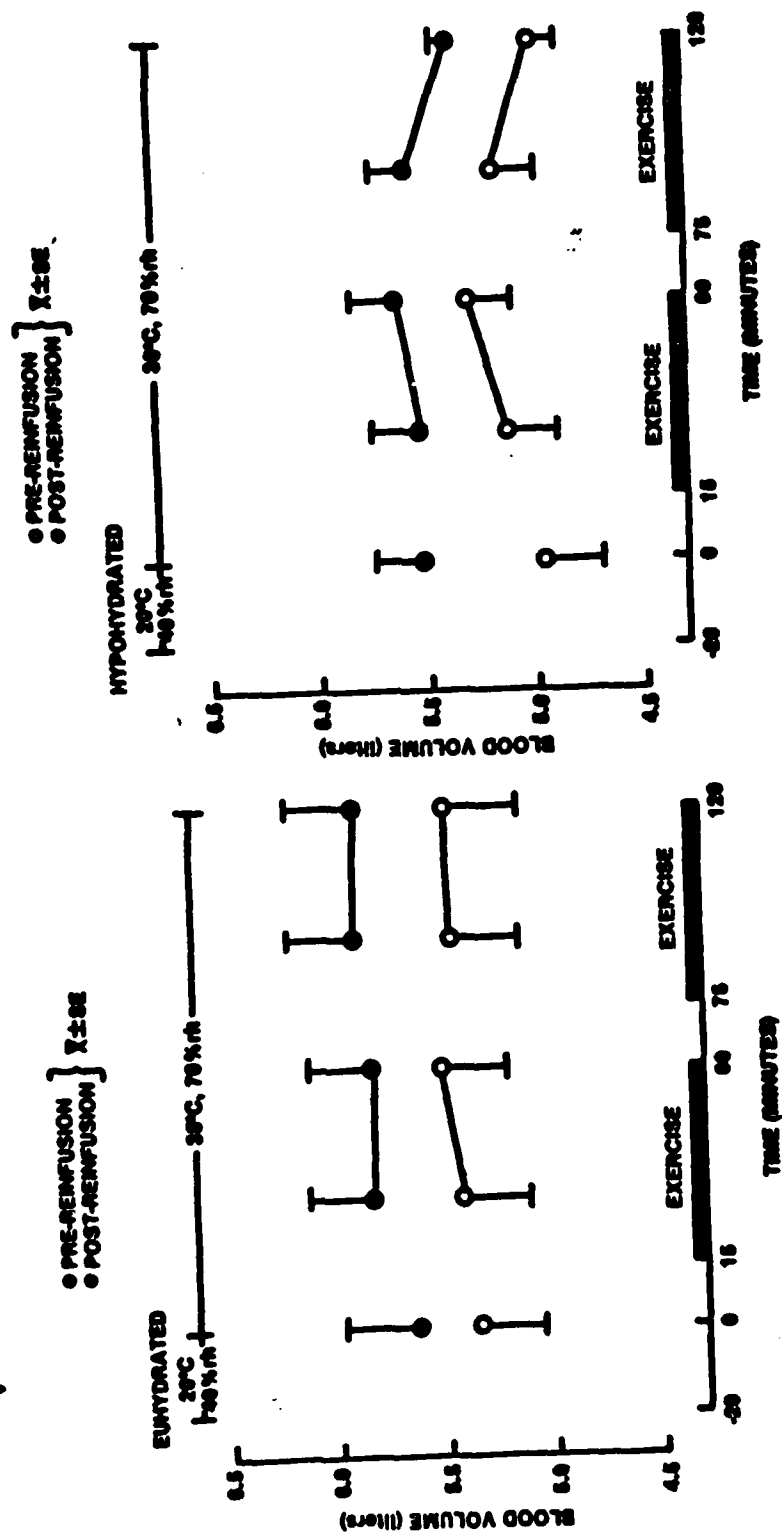


Fig. 4.

